

CLAIMS

What is claimed is:

- 1 1. A method for use in detecting the presence of a selected
2 microscopic pathogen in a sample comprising:
 - 3 (a) providing a substrate having a detection region thereon
4 comprising a surface comprising microstructures including depressions of width and
5 depth sized to align a liquid crystal material in contact therewith and wherein the
6 depressions are of a size sufficient to be occupied by the selected pathogen; and
7 (b) treating the surface of the detection region to provide a layer
8 thereon that blocks non-specific binding of pathogens to the surface and that
9 includes a binding agent that specifically binds the selected pathogen to be detected.
- 1 2. The method of Claim 1, further comprising applying a sample
2 to be tested for the presence of the specific pathogen to the surface of the detection
3 region of the substrate, and thereafter applying the liquid crystal material to the
4 detection region that will be aligned by the microstructures on the surface of the
5 substrate in the absence of binding of pathogen particles to the surface of the
6 substrate, whereby the presence of the selected pathogen in the sample will be
7 manifested by a visually observable disordering of the liquid crystal material caused
8 by the pathogen particles bound to the substrate.
- 1 3. The method of Claim 1, further comprising coating at least a
2 portion of the detection region with an inorganic material selected from the group
3 consisting of an oxide of silicon, an oxide of a metal, a metal, and combinations
4 thereof.
- 1 4. The method of Claim 4, wherein the inorganic material is
2 silver or gold and the method further comprises treating at least a portion of the
3 silver or gold with a mercaptan or a disulfide.
- 1 5. The method of Claim 1, wherein the substrate is formed of a
2 molded polymer plastic.

1 6. The method of Claim 5, wherein the molded polymer plastic
2 comprises polystyrene, polycyanoacrylate, or polyurethane.

1 7. The method of Claim 5, wherein the molded polymer is
2 polydimethylsiloxane.

1 8. The method of Claim 1, wherein the treating of the surface of
2 the detection region includes applying bovine serum albumin to the surface of the
3 detection region of the substrate.

1 9. The method of Claim 8, wherein the treating of the surface of
2 the detection region includes applying an immunoglobulin or a portion thereof to the
3 detection region surface that provides a specific binding site for the selected
4 pathogen.

1 10. The method of Claim 1, wherein the selected pathogen is a
2 virus and the depressions on the surface of the detection region have a width and
3 depth in the range of 5 nm to 500 nm.

1 11. The method of Claim 1, wherein the depressions on the
2 surface of the detection region of the substrate comprise parallel grooves having a
3 width of approximately 100 nm.

1 12. The method of Claim 11, wherein the grooves are separated
2 by ridges having a width of about 100 nm.

1 13. The method of Claim 11, wherein the grooves have a depth of
2 approximately 100 nm.

1 14. The method of Claim 1, wherein the binding agent is selected
2 from the group consisting of peptides, polypeptides, RNA, DNA, biotin, avidin,
3 fragments of antibodies, antibodies, and sugars.

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1 15. The method of Claim 1, wherein the selected pathogen is a
2 bacteria and the depressions on the surface of the detection region have a width and
3 depth in the range of 0.1 μm to 10 μm .

1 16. The method of Claim 1, wherein substantially all the binding
2 agent is located in the depressions of the detection region.

1 17. The method of Claim 1, further comprising contacting
2 magnetic beads with the sample to be tested for the presence of the specific
3 pathogen; thereafter contacting the magnetic beads with the surface of the detection
4 region of the substrate; and thereafter applying the liquid crystal material to the
5 detection region, wherein the depressions are of a size sufficient to be occupied by
6 the magnetic beads after contacting the pathogen, whereby the presence of the
7 selected pathogen in the sample will be manifested by a visually observable
8 disordering of the liquid crystal material.

1 18. The method of claim 17, wherein the magnetic beads have a
2 surface comprising a binding agent that specifically binds the pathogen to be tested.

1 19. A detection apparatus for use in the detection of the presence
2 of a selected pathogen in a sample comprising:
3 a substrate with a detection region on a surface thereof, the detection
4 region having microstructures comprising grooves formed therein that will align
5 liquid crystal material in contact therewith, the width and depth of the grooves
6 being in the range of 10 μm or less; a blocking layer on the surface of the detection
7 region of the substrate that does not disrupt the alignment of liquid crystal material
8 in contact therewith, the blocking layer blocking nonspecific adsorption of
9 pathogens to the surface; and a binding agent on the surface of the detection region
10 of the substrate, the binding agent specifically binding the selected pathogen.

1 20. The detection apparatus of Claim 19, wherein the selected
2 pathogen is a virus and the width and depth of the grooves are in the range of 5 nm
3 to 500 nm.

1 29. The detection apparatus of Claim 19, wherein the selected
2 pathogen is a bacteria and the width and depth of the grooves are in the range of 0.1
3 μm to 10 μm .

1 30. The detection apparatus of Claim 19, wherein the substrate is
2 formed of polydimethylsiloxane.

1 31. The detection apparatus of Claim 19, wherein the substrate
2 has multiple detection regions in an array on the surface of the substrate, each of the
3 detection regions having a binding agent thereon that binds a different specific
4 pathogen.

1 32. The detection apparatus of Claim 19, wherein the detection
2 region is a first detection region and the substrate further comprises at least a
3 second detection region on the surface of the substrate, the at least second detection
4 region of the substrate having microstructures comprising grooves formed therein
5 having a width and a depth that will align liquid crystal material in contact
6 therewith, wherein the width of the grooves in the at least second detection region is
7 different from the width of the grooves in the first detection region; the depth of the
8 grooves in the at least second detection region is different from the depth of the
9 grooves in the first detection region; or both the width and depth of the grooves in
10 the at least second detection region are different from the width and depth of the
11 grooves in the first detection region.

1 33. The detection apparatus of Claim 19, wherein substantially all
2 the binding agent is located in the grooves of the detection region.

1 34. A method for use in detecting the presence of a selected
2 microscopic pathogen in a sample comprising:

3 (a) providing a substrate having a detection region thereon
4 comprising a surface comprising microstructures including depressions of width and
5 depth sized to align a liquid crystal material in contact therewith and wherein the
6 depressions are of a size sufficient to be occupied by the selected pathogen, the
7 surface of the detection region treated to block non-specific binding of pathogens to
8 the surface and having a binding agent thereon that specifically binds the selected
9 pathogen to be detected;

003.275909.1

10 (b) applying a sample to be tested for the presence of the specific
11 pathogen to the surface of the detection region of the substrate; and

12 (c) thereafter applying the liquid crystal material to the detection
13 region that will be aligned by the microstructures on the surface of the substrate in
14 the absence of binding of particles of the pathogen to the surface of the substrate,
15 whereby the presence of the selected pathogen in the sample will be manifested by a
16 visually observable disordering of the liquid crystal material caused by the pathogen
17 particles bound to the substrate in the depressions.

1 35. The method of Claim 34, further comprising coating at least a
2 portion of the detection region with an inorganic material selected from the group
3 consisting of an oxide of silicon, an oxide of a metal, a metal, and combinations
4 thereof.

1 36. The method of Claim 35, wherein the inorganic material is
2 silver or gold and the method further comprises treating at least a portion of the
3 silver or gold with a mercaptan or a disulfide.

1 37. The method of Claim 34, wherein the substrate is formed of a
2 molded polymer plastic.

1 38. The method of Claim 37, wherein the molded polymer plastic
2 comprises polystyrene, polycyanoacrylate, or polyurethane.

1 39. The method of Claim 37, wherein the molded polymer is
2 polydimethylsiloxane.

1 40. The method of Claim 34, wherein the surface of the detection
2 region includes a layer of bovine serum albumin on the surface to block non-
3 specific binding of the pathogens.

1 41. The method of Claim 34, wherein the surface of the detection
2 region includes an immunoglobulin or a portion thereof on the detection region
3 surface that provides a specific binding site for the selected pathogen.

1 42. The method of Claim 34, wherein the selected pathogen is a
2 virus and the depressions on the surface of the detection region have a width and
3 depth in the range of 5 nm to 500 nm.

1 43. The method of Claim 34, wherein the depressions on the
2 detection region of the substrate comprise parallel grooves having a width and depth
3 of approximately 100 nm.

1 44. The method of Claim 34, wherein the binding agent is
2 selected from the group consisting of peptides, polypeptides, RNA, DNA, biotin,
3 avidin, fragments of an antibody, antibodies, and sugars.

1 45. The method of Claim 34, wherein the selected pathogen is a
2 bacteria and the depressions on the surface of the detection region have a width and
3 depth in the range of 0.1 μm to 10 μm .

1 46. A kit for use in the detection of the presence of a selected
2 pathogen in a sample comprising:
3 (a) a substrate with a detection region on a surface thereof, the
4 detection region having microstructures comprising grooves formed therein that will
5 align liquid crystal material in contact therewith, the width and depth of the grooves
6 being in the range of 10 μm or less, a blocking layer on the surface of the detection
7 region of the substrate that does not disrupt the alignment of liquid crystal material
8 in contact therewith, the blocking layer blocking nonspecific adsorption of
9 pathogens to the surface and a binding agent attached on the surface of the detection
10 region of the substrate, the binding agent specifically binding the selected pathogen;
11 and

12 (b) liquid crystal material that will be aligned when in contact
13 with the detection region of the substrate in the absence of pathogens bound to the
14 detection region.

1 47. The kit of Claim 46, wherein the selected pathogen is a virus
2 and the width and depth of the grooves are in the range of 5 nm to 500 nm.

1 48. The kit of Claim 47, wherein the grooves are separated by
2 ridges having a width on the order of 100 nm or less.

1 49. The kit of Claim 46, wherein at least a portion of the
2 detection region is coated with an inorganic material selected from the group
3 consisting of an oxide of silicon, an oxide of a metal, a metal, and combinations
4 thereof.

1 50. The kit of Claim 49, wherein the inorganic material is gold or
2 silver and at least a portion of the silver coated region or the gold coated region
3 comprises a reaction product of the gold or silver with a mercaptan or a disulfide.

1 51. The kit of Claim 46, wherein the substrate is formed of a
2 polymer plastic.

1 52. The kit of Claim 51, wherein the polymer plastic comprises
2 polystyrene, polycyanoacrylate, or polyurethane.

1 53. The kit of Claim 46, wherein the blocking layer is formed of
2 bovine serum albumin.

1 54. The kit of Claim 46, wherein the binding agent comprises an
2 immunoglobulin or a portion thereof which specifically binds the selected pathogen.

1 55. The kit of Claim 46, wherein the binding agent is selected
2 from the group consisting of peptides, polypeptides, RNA, DNA, biotin, avidin,
3 fragments of an antibody, antibodies, and sugars.

1 56. The kit of Claim 46, wherein the selected pathogen is a
2 bacteria and the width and depth of the grooves are in the range of 0.1 μm to 10
3 μm .

1 57. The kit of Claim 46, wherein the substrate is formed of
2 polydimethylsiloxane with the grooves molded therein.

1 58. The kit of Claim 46, wherein the liquid crystal material is 4-
2 cyano-4'-pentylbiphenyl nematic liquid crystal.

1 59. The kit of Claim 46, wherein the substrate has multiple
2 detection regions in an array on the surface of the substrate, each of the detection
3 regions having a binding agent thereon that binds a different specific pathogen.

1 60. The kit of Claim 46, wherein the detection region is a first
2 detection region and the substrate further comprises at least a second detection
3 region on the surface of the substrate, the at least second detection region of the
4 substrate having microstructures comprising grooves formed therein having a width
5 and a depth that will align liquid crystal material in contact therewith, wherein the
6 width of the grooves in the at least second detection region is different from the
7 width of the grooves in the first detection region; the depth of the grooves in the at
8 least second detection region is different from the depth of the grooves in the first
9 detection region; or both the width and depth of the grooves in the at least second
10 detection region are different from the width and depth of the grooves in the first
11 detection region.

1 61. The kit of Claim 46, wherein substantially all the binding
2 agent is located in the grooves of the detection region

1 62. The kit of Claim 46, further comprising magnetic beads of a
2 size sufficient to fit into the grooves of the detection region.

1 63. The kit of Claim 62, wherein the magnetic beads comprise a
2 surface and the surface of the magnetic beads comprise a binding agent that binds
3 the selected pathogen.